

MYCOTAXON

<http://dx.doi.org/10.5248/130.215>

Volume 130, pp. 215–221

January–March 2015

***Morchella galilaea*, an autumn species from Turkey**HATIRA TAŞKIN¹*, HASAN HÜSEYİN DOĞAN², & SAADET BÜYÜKALACA¹¹Department of Horticulture, Faculty of Agriculture, University of Çukurova,
01330 Adana, Turkey²Department of Biology, Faculty of Science, University of Selçuk
42074, Konya, Turkey*CORRESPONDENCE TO: hatirataskin1@gmail.com

ABSTRACT — *Morchella* specimens collected during the autumn seasons of 2009–12 from Adana Province, Turkey, have been identified by DNA sequencing as representatives of *Morchella galilaea*. The macro- and micromorphology, phylogeny, ecology, and fruiting phenology of the Turkish material are evaluated.

KEY WORDS — mist units, morels, taxonomy, volcanic media

Introduction

Morchella species (true morels) are some of the most valuable and important fungi consumed worldwide due to their unique aroma and flavor. Their commercial value has been increasing over the years as a result of growing consumer demand. Countries with diverse morel species include China, USA, Pakistan, India, and Turkey, which export morels to European countries (Pilz et al. 2007). In Turkey, morels generally grow in pine forests (*Pinus brutia*, *P. nigra*) from late February to June (Taşkın & Büyükalaca 2012). According to a review by Pilz et al. (2007), researchers have reported *Morchella* associated with elm, ash, aspen, tulip poplar, apple, and cottonwood trees. Although morel fruiting season occurs generally during the spring season, Sturgis (1905) and Masaphy et al. (2009) have reported morels fruiting in different seasons (Pilz et al. 2007). Morels may be found throughout Turkey, especially in Mediterranean and Aegean regions. If they can be collected in relatively large volumes, morels offer the potential of providing a significant income for people living near the forests and could be regarded as a significant export product for Turkey.

In addition to the existence of an endemic Turkish morel species, *M. anatolica* Işiloğlu et al. (Işiloğlu et al. 2010), Taşkın et al. (2010, 2012)

identified 20 phylogenetically distinct *Morchella* species in Turkey. Nineteen of these species were collected from forests in spring, but one (given the phylogenetic code *Mes-16*) was observed in autumn in the mist units of the Horticultural Research and Application Area of Çukurova University, Adana, Turkey (Taşkın et al. 2012). Based on studies by Taşkın et al. (2012), O'Donnell et al. (2011), Du et al. (2012a,b) we named this population as a new species but later realized that it represented *Morchella galilaea*, described from Israel by Clowez (2012). However, as detailed morphological, microscopical, and ecological features were not provided in the original description, we have evaluated the morphology, phylogenetic position, ecology, and fruiting phenology of *M. galilaea* collected from Turkey. Our results should be beneficial to future studies concerned with culture and conservation of *Morchella* spp.

Materials & methods

Specimens were collected from Adana province in Turkey (Horticultural Research and Application Area of Çukurova University) during autumn and early winter (October–December) between 2009 and 2012 (Fig. 1). Two (in 2009), two (in 2010), and 11 (in 2012) *Morchella* specimens were collected from mist units used for rooting agricultural plant cuttings.

Molecular analyses were performed at the United States Department of Agriculture (USDA-NCAUR-Peoria-IL) in USA (Taşkın et al. 2012). These samples were analyzed through partial RNA polymerase I (RPB1), RNA polymerase II (RPB2), translation elongation factor (EF1- α), 28S (LSU) rRNA gene and internal transcribed spacer (ITS) rRNA gene sequences. Macro- and microscopical characters were studied at the Department of Biology, Selçuk University (Konya, Turkey). The specimens were mounted in Melzer's reagent and examined microscopically using a Leica DM 750 microscope. Volcanic tuff was analyzed at Alata Horticultural Research Institute, and pH (Jackson 1962), salinity (Soil Survey Division Staff 1993), total lime (CaCO_3 ; Çağlar 1949), organic matter (Walkley & Black 1934), phosphorus (P; Olsen & Dean 1965), magnesium (Mg), calcium (Ca), iron (Fe), zinc (Zn), manganese (Mn), and copper (Cu) were analyzed using an atomic absorption spectrophotometer (Varian SpectrAA Model 220FS, Germany).

Taxonomy

Morchella galilaea Masaphy & Clowez, Bull. Soc. Mycol. Fr. 126 (3-4): 238.

2012 ["2010"].

Figs 1–2

ASCOMATA 53–78 mm tall. HYMENOPHORE 36–45 mm tall, 12.5–17.5 mm wide, conic to subcylindric, pitted and ridged, ridges have two types, these are narrow polygonal ridges and long elliptical ridges. STERILE RIBS longitudinally arranged, 5.2–9.8 mm wide, 14.5–24.74 mm tall, thin, elastic, simple, or forked and anastomosing to form elongated hymenial pits, white, bluntly rounded when young, but often flattened in age, ridges glabrous.



FIG. 1 – *Morchella galilaea* specimens collected from mist units on the Çukurova University campus (Adana, Turkey).

HYMENIAL PITS light gray, whitish gray, blackish gray or silvery gray, when young; becoming, greyish-light brown, light olive brown or yellow brown with maturity. ASCOCARP MARGIN connecting directly into the stalk without a sharp bend, free. STIPE 17–33 mm high, 5–8.2 mm wide, cylindrical and thickening slightly towards the base; surface when young completely glabrous and white, in maturity light yellowish white. CONTEXT whitish, 0.5–1 mm thick in the hollow hymenophore; sterile inner surface whitish, glabrous. ASCOPORES elliptical, smooth (surface distinctly rugulate to tectate under the scanning electron microscope), with homogeneous contents, (12.5–)13.5–18(–19.5) $\times$ (7.5–)8–9.5(–10) μm [average $Q = 1.83$]. ASCI 8-spored, 165–220 $\times$ 15–22 μm , cylindric, hyaline. PARAPHYSES cylindric to subclavate, apices rounded to subacute, with 2–3-septate, hyaline or with brown to brownish homogeneous contents in KOH (2%), 100–130 $\times$ 6–11 μm . RESIDUAL PARAPHYSES on sterile ridges similar to paraphyses, hyaline or with bundled brownish contents.

SPECIMENS COLLECTED: Turkey. Adana: Çukurova University Campus, 37°01'55"N 35°22'12"E, alt. 39 m, solitary or scattered on volcanic tuff in misted cutting propagation beds, 17 Nov 2009, leg. H. Taşkın, Taskin 117 (ANK); 28 Nov 2012, leg. H.H. Doğan, Doğan 10022 (Kon Fungarium).

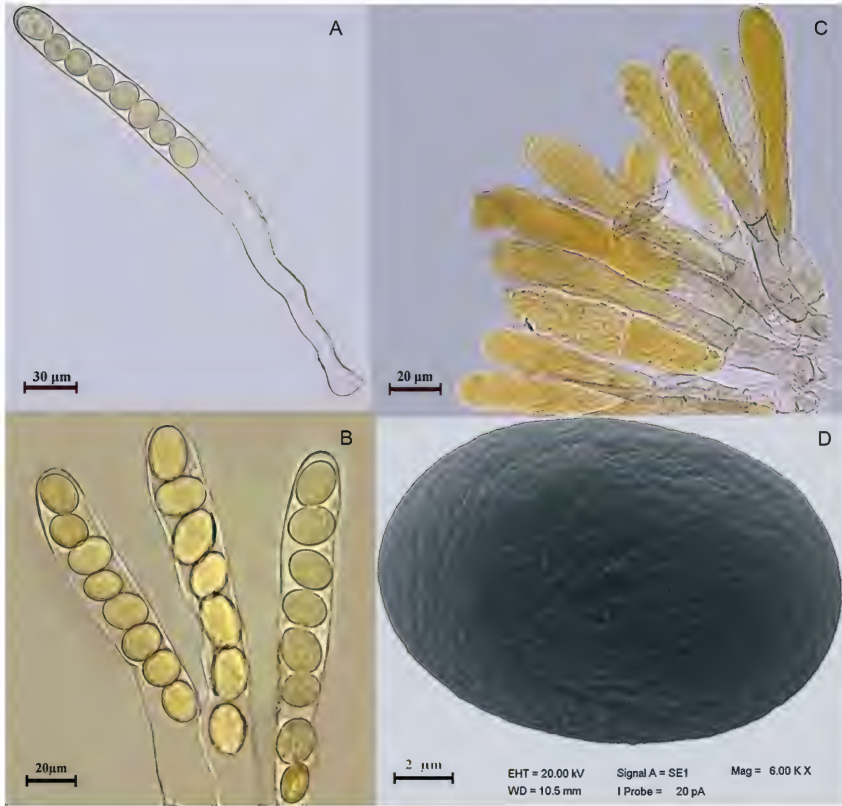


FIG. 2 – *Morchella galilaea* (Turkish specimen, Taskin 117).
A, B: asci with ascospores. C: paraphyses. D: ascospore (SEM).

Discussion

The morphological features of *M. galilaea* were cited by Clowez (2012) as: ascomata 3–5 cm height, ovoid, light yellow; primary alveolus large, few; edges of ribs same color and rusty; stipe cylindrical rather slender, attenuated at the top and wider at the base.

When we compared *M. galilaea* samples collected from Israel and Turkey morphologically, we found certain differences between the samples, which we summarize as follows: (a) ascomata of the Israel specimens were shorter (3–5 cm); (b) the stipes of the Turkish specimens were more or less cylindrical except for the slightly thicker base while in the Israeli collection the stipes had a narrower apex and wider base; (c) the Israeli specimen was found under *Fraxinus syriaca*, while our Çukurova University specimens were collected from a no tree zone.

Molecular characterization

Morchella galilaea [as *Mes-16*] has been molecularly assessed through GCPSR analyses by Taşkın et al. (2012) and also molecularly analyzed by O'Donnell et al. (2011), Du et al. (2012 a, b), Masaphy et al. (2010), Clowez (2012), and Richard et al. (2015). These studies indicate that *M. galilaea* occurs in China (Du et al. 2012a,b), Java, Hawaii, Israel (Masaphy et al. 2010, Clowez 2012), New Zealand, India (Kanwal et al. 2011), and three African countries as well as in Turkey. The Turkish and Israel sequences of *Morchella galilaea* are deposited in GenBank-ITS1-5.8S-ITS2 as JQ723082 and GU589858, respectively (Masaphy et al. 2010; Clowez 2012; Du et al. 2012 a, b).

Ecological assessment

The results of the volcanic tuff analyses are provided in TABLE 1. Relatively high levels of copper, manganese, zinc, iron, magnesium and calcium were detected in the volcanic tuff medium, which was identified as having a loam soil structure, normal CaCO_3 , and alkali salinity profile. There was a relatively low potassium level. Singh et al. (2004), who determined that pH values vary from 6.5 to 7 in more natural areas in India, detected high levels of calcium, carbon, nitrogen, sodium, and lead and low levels of phosphates, chloride, and potassium; soil type varied from sandy to loam.

Sturgis (1905) observed morels fruiting in September in an aspen-spruce forest burned the previous summer in southwestern British Columbia (Pilz et al. 2007). Masaphy et al. (2009) reported that a *Morchella* species emerged from late autumn to late spring in northern Israel under *Rhamnus alaternus*,

TABLE 1. Properties of the volcanic tuff in which *Morchella galilaea* was found fruiting.

PROPERTY	RESULTS
Texture (100 g/ml)	46
CaCO_3 (%)	19.6
Salinity (mmhos/cm)	0.06
Organic matter (%)	2.6
pH	7.6
Available potassium (ppm)	167.5
Available phosphorus (ppm)	19.2
Calcium (ppm)	2300
Available magnesium (ppm)	759
Iron (ppm)	6.6
Zinc (ppm)	2.8
Manganese (ppm)	4.8
Copper (ppm)	1.6

Platanus orientalis, *Populus euphratica*, *Laurus nobilis*, and *Fraxinus syriaca*. However, they could not find a mycorrhizal relationship between trees and morels. *Morchella* is known to be saprobic. However, according to a review study conducted by Tedersoo et al. (2010), *Morchella* species may associate weakly with plant roots and thus are likely to be facultative biotrophs. Nonetheless, some researchers believe that certain *Morchella* species may be able to form mycorrhizal relationships with certain trees. Dahlstrom et al. (2000) reported a mycorrhizal relationship between *Morchella* and species of *Pinaceae*, and Buscot & Kottke (1990) detected a mycorrhizal association between *Morchella* and *Picea abies*. Our morel specimens grew in a mist unit. While this unit was used for rooting olive cuttings during 2009–2010, no cuttings were placed into the units in 2012, which suggests that our specimens are probably not mycorrhizal. Our hypothesis is that the mycelium of the species was carried to the mist units by means of the volcanic tuff, where it developed over the years. After mycelial development, a sprinkle irrigation system and hormone application used for rooting might also have stimulated *Morchella* fruiting. The same species has been found under *Populus* and *Quercus* by Du et al. (2012b) and under *Fraxinus syriaca* by Masaphy et al. (2010). Although morels typically fruit from late February to early June, our *Morchella* specimens were collected between October and December. In 2012, the air temperature at Çukurova University-Adana averaged 29°C in October, 23°C in November, and 16°C in December.

Acknowledgements

The authors would like to express their sincere gratitude to Dr. Kerry O'Donnell (USDA-NCAUR-Peoria, IL, USA) for allowing us to perform molecular analyses in his laboratory. A special thanks goes to Dr. Davut Keleş (Manager of Alata Horticultural Research Institute), for the volcanic tuff analysis, and to Berken Çimen, for organization of the figures. We are also indebted to Prof. Dr. Mitko Karadelev, Prof. Dr. Aysun Pekşen, Prof. Dr. Daniel Royse for their critical reviews of the manuscript.

Literature cited

- Buscot F, Kottke I. 1990. The association of *Morchella rotunda* (Pers.) Boudier with roots of *Picea abies* (L.) Karst. *New Phytologist* 116: 425–430.
- Çağlar KO. 1949. Soil science. Ankara University Agriculture Faculty Publication, Ankara.
- Clowez P. 2012 ('2010'). Les morilles: une nouvelle approche mondiale du genera *Morchella*. *Bulletin de la Société Mycologique de France* 126: 199–376.
- Dahlstrom JL, Smith JE, Weber NS. 2000. Mycorrhiza-like interaction by *Morchella* with species of the *Pinaceae* in pure culture synthesis. *Mycorrhiza* 9: 279–285.
- Du XH, Zhao Q, Yang ZL, Hansen K, Taşkın H, Büyükalaca S, Dewberry D, Moncalvo JM, Douhan GW, Robert V, Crous PW, Rehner SA, Rooney AP, O'Donnell K, Sink S. 2012a. How well do ITS rDNA sequences 'barcode' species of true morels (*Morchella*)? *Mycologia* 104: 1351–1368. <http://dx.doi.org/10.3852/12-056>

- Du XH, Zhao Q, Yang ZL, O'Donnell K, Rooney AP, Yang ZL. 2012b. Multigene molecular phylogenetics reveals true morels (*Morchella*) are especially species-rich in China. *Fungal Genetics and Biology* 49: 455–469. <http://dx.doi.org/10.1016/j.fgb.2012.03.006>
- İşiloğlu M, Alli H, Spooner BM, Solak MH. 2010. *Morchella anatolica* (Ascomycota), a new species from southwestern Anatolia, Turkey. *Mycologia* 102: 455–458. <http://dx.doi.org/10.3852/09-186>
- Jackson ML. 1962. Soil chemical analysis. Prentice Hall, New York.
- Kanwal HK, Acharya K, Ramesh G, Reddy MS. 2011. Molecular characterization of *Morchella* species from the western Himalayan region of India. *Current Microbiology* 62: 1245–1252. <http://dx.doi.org/10.1007/s00284-010-9849-1>
- Masaphy S, Zabari L, Goldberg D. 2009. New long season ecotype of *Morchella rufobrunnea* from northern Israel. *Micologia Aplicada International* 21: 45–55.
- Masaphy S, Zabari L, Goldberg D, Jander-Shagug G. 2010. The complexity of *Morchella* systematics: a case of the yellow morel from Israel. *Fungi* 3(2): 14–18.
- O'Donnell K, Rooney AJ, Mills GL, Kuo M, Weber NS, Rehner SA. 2011. Phylogeny and historical biogeography of true morels (*Morchella*) reveals an early Cretaceous origin and high continental endemism and provincialism in the Holarctic. *Fungal Genetics and Biology* 48: 252–265. <http://dx.doi.org/10.1016/j.fgb.2010.09.006>
- Olsen SR, Dean LA. 1965. Estimation of available phosphorus in soils by extraction with sodium bicarbonate. United States Department of Agriculture, Washington, D.C.
- Pilz D, Mclain R, Alexander S, Villarreal-Ruiz L, Berch S, Wurtz TL, Parks CG, McFarlane E, Baker B, Molina R, Smith JE. 2007. Ecology and management of morels harvested from the forests of western North America. General Technical Report, PNW-GTR-710. United States Department of Agriculture, Forest Service, Pacific Northwest Research Station, Portland.
- Richard F, Sauve M, Bellanger JM, Clowez P, Kansen H, O'Donnell K, Urban A, Courtecuisse R, Moreau PA. 2015. True morels (*Morchella*, *Pezizales*) of Europe and North America: Evolutionary relationships inferred from multilocus data and a unified taxonomy. *Mycologia* <http://dx.doi.org/10.3852/14-166>.
- Singh SK, Kamal S, Tiwari M, Rai RD, Upadhyay RC. 2004. Myco-ecological studies of natural morel bearing sites in Shivalik Hills of Himachal Pradesh, India. *Micologia Aplicada International* 16: 1–6.
- Soil Survey Division Staff. 1993. Soil survey manual. United States Department of Agriculture Handbook 18. Soil Conservation Service.
- Sturgis WC. 1905. Remarkable occurrence of *Morchella esculenta* (L.) Pers. *Journal of Mycology* 11: 269.
- Taşkın H, Büyükalaca S. 2012. Morel (*Morchella*) mushroom. *Bahçe* 41: 25–36.
- Taşkın H, Büyükalaca S, Doğan HH, Rehner SA, O'Donnell K. 2010. A multigene molecular phylogenetic assessment of true morels (*Morchella*) in Turkey. *Fungal Genetics and Biology* 47: 672–682. <http://dx.doi.org/1016/j.fgb.2010.05.004>
- Taşkın H, Büyükalaca S, Hansen K, O'Donnell K. 2012. Multilocus phylogenetic analysis of true morels (*Morchella*) reveals high levels of endemics in Turkey relative to other regions of Europe. *Mycologia* 104: 446–461. <http://dx.doi.org/10.3852/11-180>
- Tedersoo L, May TW, Smith M. 2010. Ectomycorrhizal lifestyle in fungi: global diversity, distribution, and evolution of phylogenetic lineages. *Mycorrhiza* 20: 217–263.
- Walkley A, Black LA. 1934. An examination of the Degtjareff method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Science* 39: 29–38.